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Recurrent non-canonical histone H3 mutations in spinal cord diffuse gliomas

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Somatic mutations in the *H3F3A* and *HIST1H3B* genes encoding the histone H3 variants H3.3 and H3.1, respectively, are important genetic drivers of diffuse gliomas in both children and adults. The recurrent p.K27M mutation in either *H3F3A* or *HIST1H3B* genes is found in the majority of diffuse gliomas centered in midline structures of the central nervous system including the thalamus, brainstem, and spinal cord where it is associated with poor prognosis irrespective of histologic grade [9–11]. “Diffuse midline glioma, H3

K27M-mutant” was thus classified as a grade IV entity in the revised 2016 WHO Classification of Tumors of the Central Nervous System. In contrast, p.G34R or p.G34V mutation in the *H3F3A* gene is found in a subset of glioblastomas located in the cerebral hemispheres of adolescents and young adults and is associated with a more favorable prognosis [6, 8–10]. While the genetic landscape of supratentorial and brainstem gliomas has now been extensively characterized [9, 11], the genetic drivers of spinal cord diffuse gliomas are less understood [1]. Here we report genomic characterization of 13 spinal cord diffuse gliomas that identified recurrent

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non-canonical histone H3 mutations including *H3F3A* p.G34W and *H3F3B* p.K27I variants.

The thirteen patients (10 male, 3 female) ranged in age from 4 to 71 years at time of diagnosis (Supplementary Table 1 [Online Resource 1]). All tumors were expansile, intramedullary masses centered in the thoracic (8), cervical (4), and lumbar (1) spinal cord. All patients underwent biopsies or subtotal resections. Microscopic evaluation demonstrated diffuse astrocytic gliomas with histologic features of diffuse astrocytoma (4), anaplastic astrocytoma (4), or glioblastoma (5). Histopathologic features are summarized in Supplementary Table 2 [Online Resource 1]. Adjuvant therapy and clinical outcomes are summarized in Supplementary Table 1 [Online Resource 1].

Targeted next-generation sequencing was performed on the 13 tumors using the UCSF500 Cancer Panel as previously described [Ref. [7] and Supplementary Table 3 (Online Resource 1)]. In total, 11 of the 13 cases harbored mutations in one of the histone H3 genes. Nine tumors harbored the recurrent p.K27M mutation in the *H3F3A* gene that defines the majority of “diffuse midline glioma, H3 K27M-mutant” (Fig. 1a and Supplementary Table 4 [Online Resource 1]). Additionally, two tumors harbored p.G34W mutation in the *H3F3A* gene, a variant that has not been previously described in CNS tumors but is the defining genetic alteration in giant cell tumor of bone [2, 3, 5]. One of these p.G34W mutations was present in a histone H3 p.K27 wild-type tumor, while the other was present in *cis* (on the same allele) as the co-occurring *H3F3A* p.K27M mutation (Fig. 1b and Supplementary Fig. 1 [Online Resource 2]). One *H3F3A* wild-type tumor instead harbored a p.K27I mutation in the *H3F3B* gene, which encodes the histone H3 variant H3.3 that is identical in amino acid sequence to the protein product encoded by the *H3F3A* gene. To the best of our knowledge, somatic mutations in the *H3F3B* gene have not been previously reported in CNS tumors. Notably, p.K36M mutation in the *H3F3B* gene is found in the vast majority of chondroblastomas [3, 5]. Additionally, this is the first example of a diffuse midline glioma harboring p.K27I mutation in a histone H3 gene instead of the more common p.K27M. The known functional consequence of the p.K27M mutation in the histone H3 genes is to block the methylation that occurs at this residue on the histone H3 tail, thus preventing this critical post-translational modification essential for promoting the transcriptional program that specifies glial differentiation [4]. As isoleucine is incapable of being methylated, this non-canonical mutation would also be expected to block methylation at this residue. To investigate the effect of this *H3F3B* p.K27I mutation, we performed

immunohistochemical staining for histone H3 lysine 27 trimethylation (H3K27me3) and found loss in the majority of tumor nuclei (Fig. 1b).

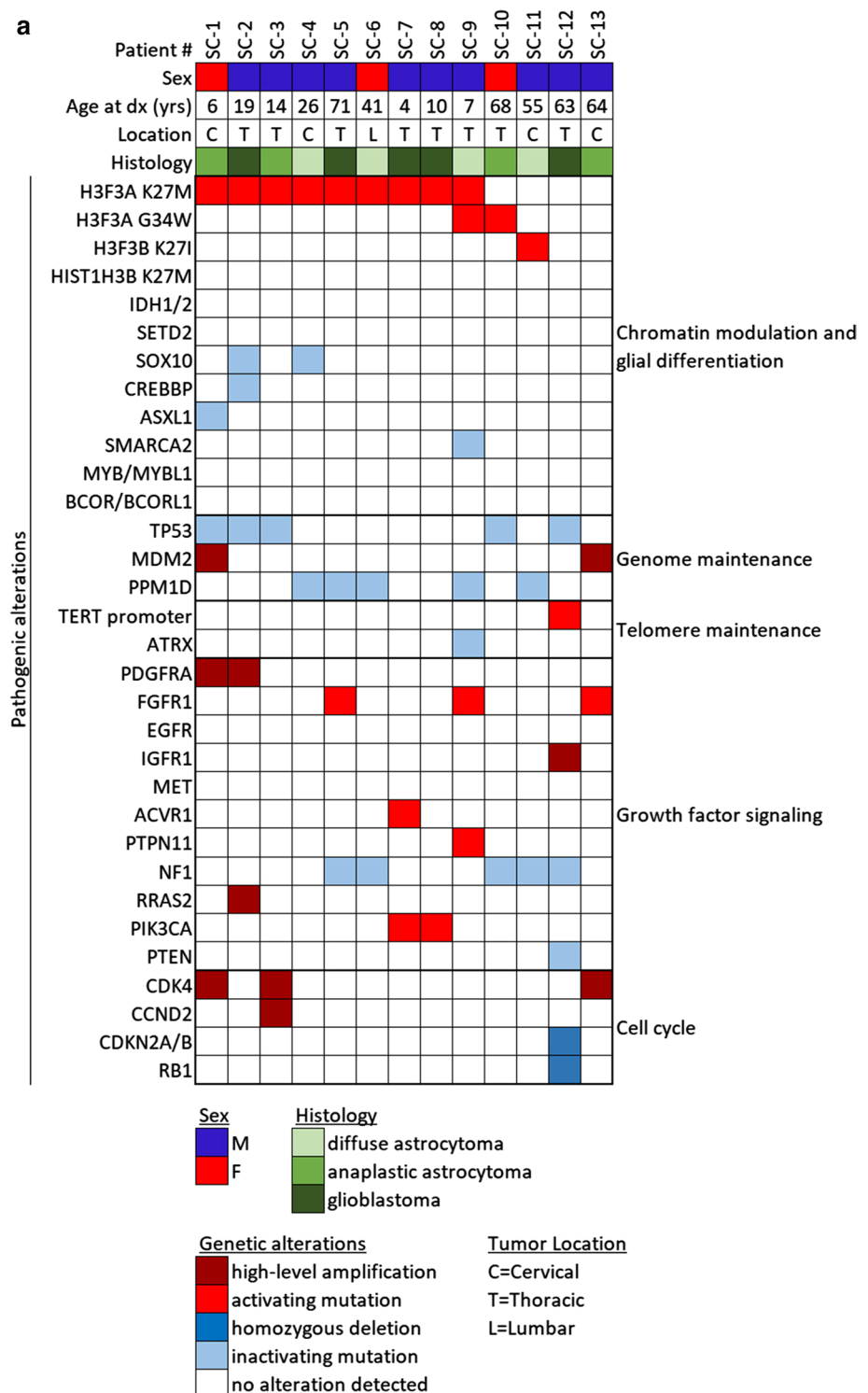
Accompanying alterations in the 11 histone H3-mutant tumors included inactivating *TP53* mutations (5) or truncating mutations in exon 6 of *PPM1D* (5) that were mutually exclusive (Fig. 1a). Two tumors harbored focal *PDGFRA* amplification, while two others had hotspot missense mutations in the kinase domain of *FGFR1* (Supplementary Table 5 [Online Resource 1]). Four tumors harbored inactivating mutations in *NF1*, another had focal *RRAS2* amplification, and another had an activating *PTPN11* mutation. Interestingly, two tumors harbored truncating frameshift mutations in the *SOX10* gene, which encodes a transcription factor important for specifying glial differentiation. While *SOX10* has not been previously described as a recurrently mutated gene in gliomas, the presence of multiple tumors in this cohort harboring truncating mutations in this gene is intriguing and warrants further investigation as to the possible role of *SOX10* mutations in gliomagenesis. While *ATRX* mutations or deletions are frequent in thalamic diffuse gliomas with H3 K27M mutation and in cerebral glioblastomas with H3 G34R/V mutation [8, 9], only 1 of the 11 histone H3-mutant spinal cord diffuse gliomas in this cohort harbored *ATRX* mutation and all were *TERT* promoter wild type.

Two of the spinal cord diffuse gliomas were histone H3 wild-type including the *H3F3A*, *HIST1H3B*, *HIST1H3C*, and *H3F3B* genes. One of these tumors had an activating *FGFR1* kinase domain mutation along with focal *MDM2* and *CDK4* amplifications, while the other harbored multiple pathogenic alterations including *CDKN2A/B* and *RB1* homozygous deletions, focal high-level *IGF1R* amplification, and *TP53*, *NF1*, *TERT* promoter, and *PTEN* mutations. Notably, none of the 13 spinal cord diffuse gliomas in this cohort were found to harbor pathogenic variants or rearrangements involving the *IDH1*, *IDH2*, *BRAF*, *RAF1*, *CIC*, *FUBP1*, *SETD2*, *MYB*, or *MYBL1* genes.

In summary, we report a series of 13 spinal cord diffuse gliomas that harbor recurrent non-canonical histone H3 mutations in a subset of cases, including *H3F3A* p.G34W and *H3F3B* p.K27I. The effect of these non-canonical histone H3 mutations on clinical outcomes has yet to be determined. Based on these findings, lack of immunoreactivity for H3 K27M-mutant protein is not sufficient to classify a diffuse glioma of the spinal cord as histone H3 wild type, and additional molecular evaluation for alternative histone H3 mutations should be considered in such cases.

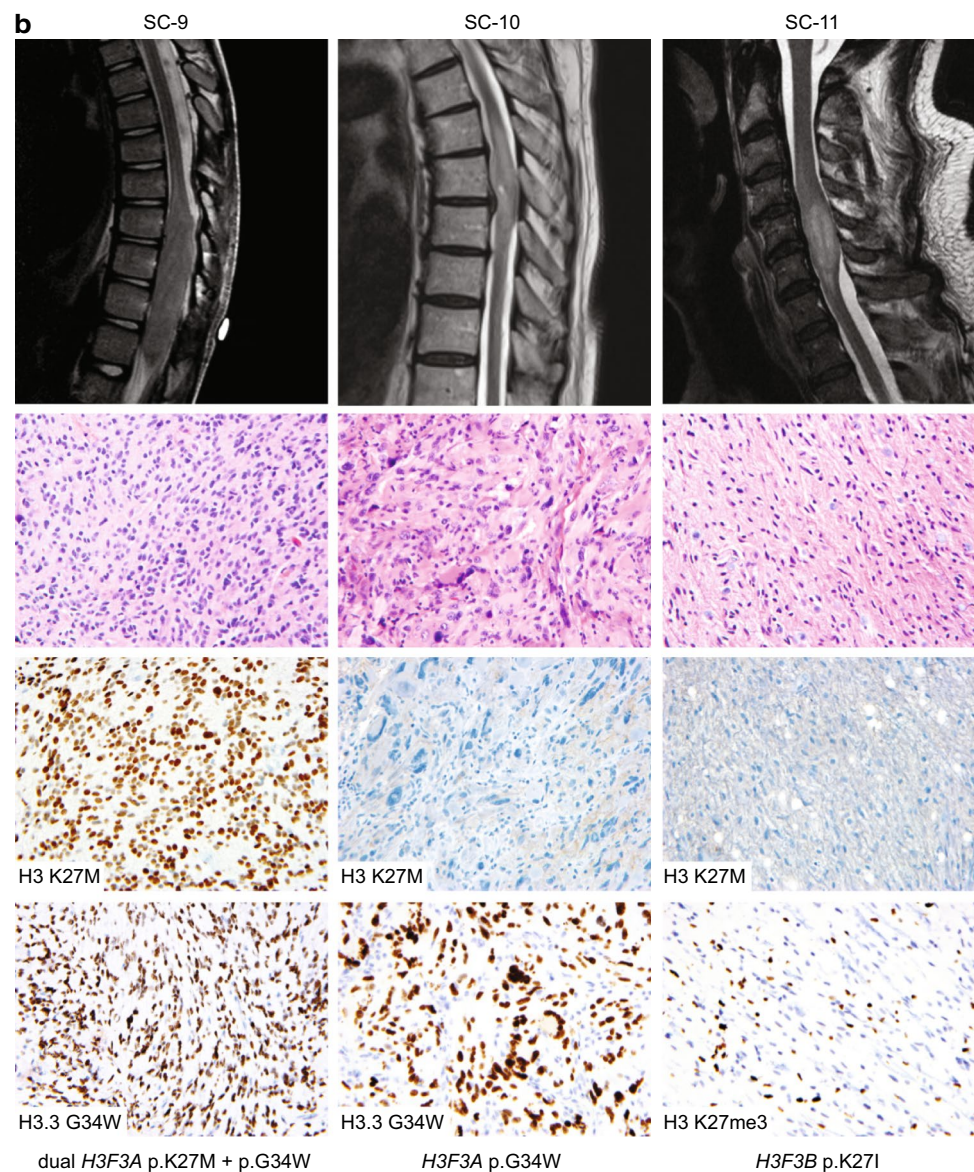
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Fig. 1 Recurrent non-canonical histone H3 mutations in spinal cord diffuse gliomas. **a** Onco-print summary table showing the clinicopathologic features and likely pathogenic genetic alterations identified in the 13 patients. **b** Radiographic and histologic features of the three spinal cord diffuse gliomas with non-canonical histone H3 mutations. Case SC-9 harboring dual *H3F3A* p.K27M and p.G34W mutations demonstrated immunopositivity with antibodies against both histone H3 K27M-mutant protein and histone H3.3 G34W-mutant protein in virtually all tumor cells. Case SC-10 harboring *H3F3A* p.G34W mutation demonstrated immunopositivity for histone H3.3 G34W-mutant protein but not histone H3 K27M-mutant protein. Case SC-11 harboring *H3F3B* p.K27I mutation was immunonegative for histone H3 K27M-mutant protein but demonstrated loss of histone H3 lysine 27 trimethylation (H3K27me3) in the vast majority of tumor nuclei



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Fig. 1 (continued)



Data availability Mutation and copy number data are available in the electronic supplementary material. Sequencing data files are available from the authors upon request.

Compliance with ethical standards

Conflict of interest The authors declare that they have no competing interests related to this report.

Ethical approval This study was approved by the Committee on Human Research of the University of California, San Francisco, with a waiver of patient consent.

References

1. Alvi MA, Ida CM, Paolini MA, Kerezoudis P, Meyer J, Barr Fritcher EG et al (2019) Spinal cord high-grade infiltrating gliomas in adults: clinico-pathological and molecular evaluation. *Mod Pathol*. <https://doi.org/10.1038/s41379-019-0271-3> (Epub 2019 Apr 26)
2. Amary F, Berisha F, Ye H, Gupta M, Gutteridge A, Baumhoer D et al (2017) H3F3A (Histone 3.3) G34W immunohistochemistry: a reliable marker defining benign and malignant giant cell tumor of bone. *Am J Surg Pathol* 41:1059–1068
3. Behjati S, Tarpey PS, Presneau N, Scheipl S, Pillay N, Van Loo P et al (2013) Distinct H3F3A and H3F3B driver mutations define chondroblastoma and giant cell tumor of bone. *Nat Genet* 45:1479–1482
4. Chan KM, Fang D, Gan H, Hashizume R, Yu C, Schroeder M et al (2013) The histone H3.3K27M mutation in pediatric glioma reprograms H3K27 methylation and gene expression. *Genes Dev* 27:985–990

5. Cleven AH, Hocker S, Briaire-de Bruijn I, Szuhai K, Cleton-Jansen AM, Bovee JV (2015) Mutation analysis of H3F3A and H3F3B as a diagnostic tool for giant cell tumor of bone and chondroblastoma. *Am J Surg Pathol* 39:1576–1583
6. Gessi M, Gielen GH, Hammes J, Dorner E, Muhlen AZ, Waha A et al (2013) H3.3 G34R mutations in pediatric primitive neuroectodermal tumors of central nervous system (CNS-PNET) and pediatric glioblastomas: possible diagnostic and therapeutic implications? *J Neurooncol* 112:67–72
7. Kline CN, Joseph NM, Grenert JP, van Ziffle J, Talevich E, Onodera C et al (2017) Targeted next-generation sequencing of pediatric neuro-oncology patients improves diagnosis, identifies pathogenic germline mutations, and directs targeted therapy. *Neuro-Oncol* 19:699–709
8. Korshunov A, Capper D, Reuss D, Schrimpf D, Ryzhova M, Hovestadt V et al (2016) Histologically distinct neuroepithelial tumors with histone 3 G34 mutation are molecularly similar and comprise a single nosologic entity. *Acta Neuropathol* 131:137–146
9. Mackay A, Burford A, Carvalho D, Izquierdo E, Fazal-Salom J, Taylor KR et al (2017) Integrated molecular meta-analysis of 1000 pediatric high-grade and diffuse intrinsic pontine glioma. *Cancer Cell* 32:520–537
10. Sturm D, Witt H, Hovestadt V, Khuong-Quang DA, Jones DT, Konermann C et al (2012) Hotspot mutations in H3F3A and IDH1 define distinct epigenetic and biological subgroups of glioblastoma. *Cancer Cell* 22:425–437
11. Wu G, Diaz AK, Paugh BS, Rankin SL, Ju B, Li Y et al (2014) The genomic landscape of diffuse intrinsic pontine glioma and pediatric non-brainstem high-grade glioma. *Nat Genet* 46:444–450

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